Residual heparin concentrations in salvaged blood from the Sorin Xtra® autotransfusion system during cardiac surgery

By

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As part of the fulfilment of requirements for the degree

Master of Medicine (MMed) in Anaesthesiology (Anes)

Faculty of Medicine and Health Sciences

University of Stellenbosch

December 2016

Topic:	**Residual heparin concentrations in salvaged blood from the Sorin Xtra® autotransfusion system during cardiac surgery**
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Abstract

Introduction

Cell salvaging is a fundamental component of blood conservation during cardiac surgery. It entails intra-operative scavenging, washing and collection of a patient's blood for retransfusion at completion of surgery. During surgery vast amounts of heparin is administered to avoid fatal thrombosis in both the bypass and autotransfusion circuits. Adequate heparin removal during the cell salvaging process is thus crucial to avoid retransfusion of heparin to these high risk patients. We wanted to measure heparin levels in the collected reinfusate prior to retransfusion, in order to quantify heparin removal in our current autotransfusion system, the Sorin Xtra®.

Method

This study was subjected to ethical committee approval prior to initiation (S14/03/050). 31 adult patients undergoing on pump cardiac surgery in Tygerberg Academic Hospital were recruited after taking informed consent. A standard cell salvaging process was used for setup using the Sorin Xtra® autotransfusion system. After completion of the cell salvaging process, a blood sample was aspirated from the collected reinfusate and stored in a standard citrated blood sampling tube. Sample processing and heparin measurement were performed in the haematology laboratory of Tygerberg Hospital. A modified anti-Xa heparin assay was employed to measure reinfusate heparin levels, since the absence of coagulation factors necessitates the addition of a set amount of normal pooled plasma prior to performing the assay.

Results

The mean heparin levels measured in the Sorin Xtra® reinfusate were 0.52 (IQR 0.16 – 0.74; 95% CI 0.30 – 0.66) IU/ml. The 95% confidence intervals did not encroach on the alternative hypothesis, but did span the value representing the null hypothesis. The data thus failed to reject the null hypothesis, indicating clinically significant reinfusate heparin levels. Sixteen of 31 reinfusates (56%; 95% confidence interval 35 to 68%) exhibited heparin concentrations exceeding 0.5 IU/ml. There was no clinically

significant relationship (r^2 0.02, p = 0.46) between heparin dosage administered to the patient and the concentration measured in the reinfusate.

Conclusion

Clinically significant heparin levels in cell saving reinfusate can potentially worsen postoperative bleeding in cardiac surgery. The mean heparin level measured in our study was more than the AABB's recommended value of 0.5IU/ml, and 16 samples had absolute values more than this. Although the absolute heparin dose retransfused remain debatably low, the possibility of heparin induced coagulopathy should be entertained in cardiac patients that received reinfusate from the Sorin Xtra® ATS with ongoing postoperative bleeding in our institution. A practical suggestion in these cases would be to quantify heparin activity either with a point of care device (TEG/ROTEM) or direct measurement of heparin concentration using an Anti-Xa assay and titrating heparin reversal accordingly.

Opsomming

Inleiding

Eritrosiet-suiweringstoestelle speel ‘n belangrike rol in bloed besparing tydens opehart chirurgie. Dit behels die versameling, was en prosessering van ‘n pasïent se eie bloed gedurende chirurgie, met die doel om die geprosesseerde bloed op ‘n latere stadium terug te transfuseer. Tydens opehart chirurgie word heparien gebruik om stolling in die omleiding- en bloedbesparingsisteme te voorkom, aangesien blootstelling aan die negatiewe oppervlakte van díe sisteme wydverspreide bloedstolling aktiveer. Heparien verwydering gedurende bloedsuiwering is dus noodsaaklik om die hertransfusie daarvan te beperk, omdat betekenisvolle heparienvlakke ‘n koagulopatie kan teweegbring in die onmiddelike postoperatiewe periode wanneer stolling van uiterste belang is. Heparien suiwering in die betrokke toestelle wat in ons instansie gebruik word is nog nie voldoende nagevors nie. Met hierdie studie het ons dus gepoog om die heparien vlakke in geprosesseerde bloed van ons huidige selsuiweringstoestel, die Sorin Xtra®, te bepaal.

Metode

Die studie is uitgevoer in Tygerberg Akademiese Hospitaal na goedkeuring van die etiese komitee(S14/03/050). 31 volwasse pasïente wat opehart chirurgie ondergaan het is gewerf na ingeligte toestemming verkry is. Slegs gevalle wat kardiopulmonale omleiding benodig het en waarvoor die Sorin Xtra® toestel gebruik is, is genader. Die bloedsuiwerings opstelling was gestandardiseer. ‘n Bloedmonster is geneem vanuit die geprosesseerde bloed na voltooiing van die suiweringsproses en in ‘n standard sitraatbuis geplaas. Die monsters is geneem na Tygerberg Hospitaal se hematologie en stollingslaboratorium vir prosessering en meting van heparienvlakke. Om die monsters se heparien vlakke akkuraat te meet is ‘n gemodifiseerde anti-Xa toets gebruik. Die afwesigheid van stolfaktore en ATIII in die geprosesseerde bloedmonsters vereis die toevoeging van normale gepoelde plasma voor die toets uitgevoer kan word.

Resultaat

Gemiddelde heparien vlakke van 0.52 (IQR 0.16 – 0.74; 95% CI 0.30 – 0.66) IU/ml is gemeet in die geprosesseerde bloed van die Sorin Xtra® toestel. Die 95% sekerheidsinterval het nie die alternatiewe hipotese se waarde ingesluit nie, maar wel die nul hipotese. Die data het nie die nul hipotese verwerp nie, wat aandui dat die gemiddelde heparien vlakke wat gemeet is wel statisties noemenswaardig is. 16 van die 31 monsters (56%; 95% sekerheidsinterval 35 to 68%) het heparien vlakke gehad van meer as 0.5IU/ml. Daar is geen kliniese verwantskap (r^2 0.02, p = 0.46) tussen die gemete heparien vlakke en die hoeveelheid heparien wat toegedien is nie.

Samevatting

Noemenswaardige heparienvlakke in geprosesseerde bloed van selbesparingstoestelle kan post operatiewe bloeding vererger na opehart chirurgie. Die gemiddelde heparien vlak in geprosesseerde bloed van die Sorin Xtra® selbesparingstoestel is meer as die AABB se voorgestelde waarde van 0.5IU/ml. Sestien van die 31 monsters het hoër vlakke gehad as die voorgestelde waarde van 0.5IU/ml. Alhoewel die absolute dosis heparien wat teruggegee word debateerbaar min is, moet heparien geinduseerde koagulopatie steeds uitgeskakel word as ‘n moontlike oorsaak van bloeding in hierdie groep pasïente wat geprosesseerde bloed van die Sorin Xtra® toestel ontvang het. Heparien aktiwiteit kan gekwantifiseer word met behulp van tromboelastografie (TEG/ROTEM) of direkte heparien vlak bepaling deur middel van ‘n anti-Xa toets om sodoende heparien omkering te lei.

Acknowledgements

I would like to acknowledge all the key personnel involved in the execution of this research.

- Marieta du Plessis from the National health laboratory services for her support as laboratory technologist.
- Mr J Harvey and Professor JF Coetzee for their support in the statistical planning and analysis of the data obtained.
- Ms A Muller, Mr H Engelbrecht and Mr M Thompson – the team of perfusion technologists from the cardiothoracic theatre complex in Tygerberg Hospital for their role in data capturing and sample collection.

I would like to thank both my promotors, Professor AI Levin and Dr WF Buys for their motivation, determination and support. They played a pivotal role in the conception and planning of this project, as well as the final drafting of the published article.

Thank you to the Harry Croxley fund for their generous research bursary.

I would lastly like to acknowledge and thank my husband Willem for his endless patience, motivation and endurance.

Declaration

I hereby declare that this document was prepared by myself, with the guidance of both my promotors, Dr WF Buys and Prof AI Levin.

This study topic was inspired by a previous similar study, conducted by Dr WF Buys and Prof AI Levin at Tygerberg Hospital. The methodology and statistical planning were kept similar as to enable comparison between the two studies for publication purposes. Both studies' results were subsequently prepared as an article, which has been published in the Journal of Cardiothoracic and Vascular Anesthesia(1).

A research grant of R15000 as granted by the Harry Croxley fund was directed to the laboratory costs involved in the research.

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Dr Mathilde Buys

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Glossary of acronyms and definitions

Acronyms

2,3 DPG:	2,3 Diphosphoglycerate
ACT:	Activated clotting time
ATP:	Adenosine triphosphate
ATIII:	Antithrombin III
ATS:	Autotransfusion system
CABG:	Coronary artery bypass grafting
CEO:	Chief executive officer
CI:	Confidence interval
CPB:	Cardiopulmonary bypass
FXa:	activated Factor X
IQR:	Interquartile range
IU:	International units
IU/ml:	International units per millilitre
MRC:	Medical Research Council
NHLS:	National Health Laboratory Services
NPP:	Normal pooled plasma
ROTEM:	Rotating thromboelastometry
TEG:	Thromboelastography

Definitions

An *autotransfusion system (ATS)* is also known as a cell saving device or cell salvage device. It entails the device and circuit used during the cell salvaging process.

The terms *"cell saving"* and *"cell salvaging"* are synonymous. They define the process where a patient's own blood is scavenged, washed, collected and retransfused to the same patient at a later stage, usually throughout the perioperative process.

The terms "*salvaged blood*", "*cell saved blood"* and "*reinfusate"* implies the completed processed product from an autotransfusion device. These terms are used interchangeably in this protocol.

"*Heparin concentration*" and "*heparin activity*" are used interchangeably in this protocol, the latter being the term that better describes heparin measurement techniques.

1. Introduction

1.1 Cell salvaging in cardiac surgery

Cell salvaging is the intraoperative process where a patient's own blood is aspirated from the surgical site, after which it is filtered, washed and suspended in normal saline for retransfusion at a later stage. An autotransfusion system (ATS) executes this process. Cardiac surgery is associated with massive blood loss, necessitating a blood conservation strategy. Autotransfusion systems are thus routinely employed in these surgeries, with the aim of minimising homologous blood transfusion. After discontinuation of cardiopulmonary bypass, the ATS processes residual blood from the bypass circuit with aspirated blood from the surgical field, after which it is retransfused to the patient. At this stage of the operation, normal coagulation is paramount to ensure proper haemostasis. This process is complicated by the fact that large doses of heparin are necessary to avoid fatal thrombosis during on-pump surgeries. The exposure of blood to the negative surface area of the bypass and cell salvage circuits results in widespread activation of the clotting cascade via the intrinsic pathway. Adequate heparin removal by the autotransfusion system is thus critical, since significant residual heparin levels in salvaged blood can contribute to inadequate haemostasis and increased postoperative bleeding.

The rationale behind the use of salvaged blood in cardiac procedures is twofold; limiting homologous blood transfusion and improved oxygen carrying capacity of reinfusate. Apart from the well known risks related to transfusion of homologous blood and blood products(2, 3) the majority of studies(4-8), barring one(9), has indicated that outcome after cardiac surgery is worse after homologous transfusion. It is therefore advisable to limit the exposure of patients to blood and blood products**.** Red cell salvaging is a fundamental component of blood conservation.(2) A 2010 Cochrane Collaboration systematic review analysed and compared results of 75 trials. The systematic review concluded that the use of cell salvaging *"reduced the rate of exposure to allogeneic red blood cell transfusion by a relative 38%"*.(10)

Salvaged blood has an improved oxygen carrying capacity when compared to homologous blood. The pH, potassium, adenosine triphosphate (ATP) and 2,3-dishosphoglycerate (2,3-DPG) levels are closer to physiological values than that of

homologous blood, optimising the position of the oxygen-hemoglobin dissociation curve.(2, 11, 12) The erythrocyte population of salvaged blood maintains their biconcave shape and constitutes the young supple cells that survived centrifugation. This improves the surface area available for oxygen dissociation across the erythrocyte membrane.

1.3 Coagulopathy, bleeding and cell salvage

Although cell salvaging during cardiac surgery has shown to decrease homologous transfusion, its effect on postoperative bleeding during cardiac surgery is less clear. Some studies show a reduction or no difference in postoperative blood loss (13-16), which is contradicted by others that show an increase in blood loss and blood product administration associated with cell salvage blood transfusion.(17) Some authors even reported an association between the volume of reinfusate administered and bleeding.(18, 19)

There are interesting interactions between hemodilution, coagulopathy and cell salvage, with some evidence that the product of cell salvage may induce a coagulopathy.(2, 18, 20, 21) The reasons why the administration of salvaged blood may induce a coagulopathy include:

1. The autotransfused “blood” is devoid of coagulation factors, fibrinogen and platelets (essentially erythrocytes suspended in a balanced salt solution). Rapid administration thereof can aggravate the existing cardiopulmonary bypass induced coagulation factor dilution.(18)
2. Platelet activation during the cell salvage process can cause transient thrombocytopaenia after reinfusion.(20)
3. Activation of coagulation by red cell fragments or by mediators released by platelets and leukocytes is a probable cause of coagulopathy. Autologous erythrocyte stroma can act as thromboplastin, initiating disseminated coagulopathy in animal models. This remains a theoretical risk as is yet to be investigated in a clinical setting.(22)
4. Significant amounts of heparin in the reinfusate could aggravate an existing coagulopathy.

5. Both heparin rebound(21, 23-25) and also rapid protamine clearance can possibly aggravate a (heparin-induced) coagulopathy.(24-32)

2. Literature review

2.1 Other devices studied

A number of studies have investigated whether the end products of ATS contain clinically significant amounts of heparin. These mostly suggest that clinically insignificant amounts of heparin remain in the end product of the Dideco Electa®, Sorin Xtra® and other autotransfusion systems(1, 33-46). These results are summarized in Table 1. These studies are subject to criticism and clinically related concerns:

1. Poorly defined heparin removal:

Heparin elimination is described qualitatively with terms like '*adequate removal*', '*virtual elimination*' and '*contains insignificant amounts'* in many of the studies. This makes them of uncertain clinical relevance since absolute heparin concentrations are not described. Others report heparin elimination as a percentage and not as absolute amounts. While elimination rates of '98.8-99.9%' and '98.7 ± 0.2%' appear impressive, these are of unknown clinical significance.

2. They are irrelevant to our institution:

The devices studied that reported absolute heparin concentrations are different from the cell saving devices employed at our institution. These studies include the ones conducted by Ottesen, Paravicini, Sandmann, McShane, Kling, Sistino, Gravlee, Rougé, Kalra, Vorweg, Geiger as stated in Table 1.

3. The accuracy of their heparin measurement is debatable:

The measurement of heparin in plasma relies on the presence of ATIII. Reinfusate, being completely devoid of any coagulation factors, has to undergo reconstitution with plasma prior to the measurement of the anti-Xa assay (see "Methodology"). The majority of published studies either did not specify their assay, and when they did specify, they did not mention whether they reconstituted their samples with NPP prior to performing the assay. This makes the accuracy doubtful. The various assays employed for each study is specified in Table 1.

In summary, the available research above suggests that no significant amounts of heparin remain in salvaged reinfusate, although their measurement technique employed is doubtful. Details of the cell saving devices employed at our institution are unknown.

2.2 Devices used at Tygerberg Hospital

The former cell saving device employed at our cardiac suites was the Dideco Electa® autotransfusion system. An initial, similar study was performed on this device in 2010 by Dr WF Buys and Prof AI Levin to determine reinfusate heparin levels(1). This study concluded that reinfusate from the Dideco Electa® device contained a mean heparin value of 0.2IU/ml(±0.17IU/ml), which was statistically insignificant. In the meantime a newer ATS, Sorin Xtra®, was employed at Tygerberg Hospital, claiming an improved heparin elimination rate. The bowl sizes used at Tygerberg Hospital are 225ml (heparin elimination of 98.8% with a standard deviation of ± 1%) and 175ml (heparin elimination of 99.9% with a standard deviation of ± 0.5%). The standard wash cycle is delineated as P_{OPT}, the default settings of the Sorin Xtra® ATS.(47) Absolute reinfusate heparin levels for these specific cycles are yet to be published.

2.4 Study objective and statistical planning

The primary objective of this study was to determine absolute heparin levels in the Sorin Xtra® ATS reinfusate in a single centred, prospective, non-randomized, observational study. The secondary objective of this study was to compare heparin concentrations in the Dideco Electa® and Sorin Xtra® reinfusates, incorporating the data obtained from the previous study performed by Buys and Levin et al. The results from both studies were reported in a research article, which has subsequently been published in the Journal of Cardiothoracic and Vascular Anesthesia.(1)

Mr Justin Harvey from the Centre for Statistical Consultation at the Tygerberg Campus, as well as professors JF Coetzee and AI Levin from the Department of Anaesthesiology and Critical Care were consulted on the design and statistical analysis of this study.

Null hypothesis: Cell salvaged blood produced by the Sorin Xtra® autotransfusion system contains clinically significant concentrations of heparin.
Alternative hypothesis: Cell salvaged blood produced by the Sorin Xtra® autotransfusion system contains clinically insignificant concentrations of heparin.

To determine how many patients would need to be studied, we interrogated published studies describing residual heparin levels in cell saver blood. The American Association of Blood Bank's 5th edition (2013) of "Standards for preoperative autologous blood collection and administration" agreed that heparin levels of less than 0.5 anti Xa IU/ml were insignificant.(48) Buys and Levin found heparin levels of 0.2 ± 0.17IU/ml in salvaged blood from the Dideco Electa® autotransfusion system(1). We thus based our analysis on finding a difference (effect size) of 0.5 IU between the null hypothesis and the alternative hypothesis. PASS (Hintze, J. (2008). PASS 2009. NCSS, LCC. Kaysville, Utah. www.ncss.com) was used to estimate the sample size. A sample size of 29 achieves 81.4% power to detect a difference of -0.5 between the null hypothesis correlation of 0.0 and the alternative hypothesis correlation of 0.5 using a two sided hypothesis test with a significance level of 0.05. This is the similar power analysis used to determine the sample size in Buys and Levin's previous study. In order to compare this study's results with that of the previously performed study, the study design, methodology and measurement techniques were kept similar. The study was performed in the same theatre complex, with a similar case mix. Heparin levels measured in the cell saver blood from the Sorin Xtra® ATS were subsequntly entered into an Excel spread sheet (Appendix A) and statistically analysed.

3. Methodology

3.1 Patient recruitment

The following inclusion criteria qualified patients to be eligible for recruitment:

I. Patients undergoing elective or emergency cardiac surgery at Tygerberg Academic Hospital where the Sorin Xtra® autotransfusion system is used for intraoperative blood salvage.
II. Patients scheduled for both coronary artery bypass graft (CABG) and/or valve replacement procedures.
III. Patients that are scheduled to have on-pump surgery.
IV. Adult patients more than 18 years of age that weigh more than 45 kilograms.
V. Patients that have agreed to participate and have given informed consent.

Exclusion criteria include

I. Paediatric cases and patients younger than 18 years of age.
II. Patients that do not give consent.
III. Non-cardiac surgery where the cell saver is going to be used.
IV. Patients that are scheduled to have off-pump surgery will be excluded from this study.

3.2 Cell salvaging process

The standard cell salvaging process employed is delineated below:

1. During the procedure, shed blood is aspirated, filtered and collected into the cell saver reservoir. After discontinuation of CPB, any remaining blood from the heart-lung machine is also processed by the ATS.
2. The aspirated fluid undergoes continuous centrifugation to separate erythrocytes from other components such as plasma, saline, platelets, proteins, free haemoglobin, amniotic fluid, fat droplets and malignant cells.
3. Once a sufficient volume has been collected, the red blood cells are washed. Salvaging setup was standardised with regards to bowl size and washing cycle. One bowl size (225ml) and wash program (P_{OPT}) were used for all cases.
4. A separate bag of normal saline without anticoagulant is used for 'washing' purposes. The reinfusate consists of salvaged erythrocytes suspended in normal saline with a haematocrit approaching 58-63%, depending on the bowl size and

wash cycle employed (bowl sizes 225 ml and 175ml are associated with a haematocrit of 63% and 58% respectively when the default washing program, P_{OPT} is used).(47)

5. The reinfusate must be transfused to the patient within 6 to 8 hours after completion of the washing process.(2)

3.3 Heparin sources

Prior to institution of cardiopulmonary bypass, the anaesthetist administers a dose of 200-300 IU/kg body weight intravenously, aiming for an ACT of more than 400 seconds. Additional heparin is administered if this target is not reached. The perfusionist adds heparin to the priming solution of the circuit during the initial setup. Additional heparin will be added during bypass time to maintain an ACT of more than 400 seconds. During cell salvaging, a solution of one litre of normal saline, to which 25 000 IU of unfractionated heparin has been added, is continuously aspirated into the cell saver reservoir to prevent clotting of the scavenged blood. Heparin levels up to 22.9 U/ml (median of 6.3 U/ml)(11) has been measured in the reservoir. This emphasises the importance of adequate heparin elimination by the ATS.

3.4 Sampling management and processing

After completion of the cell salvaging process, reinfusate is collected in a bag supplied by the manufacturer. A 4.5 millilitre sample was taken directly from the bag of reinfusate before initiation of transfusion. This sample was taken in a standard sodium citrate sampling tube and dispatched to the haematology laboratory, where it was centrifuged at 4000 revolutions per minute for 20 minutes. Sample storage at -80 degrees Celsius after centrifugation, ensures heparin stability for a considerable time. The tubes were de-identified and a consecutive numbering system employed to protect personal information of each patient. Each sample number was used to label all specimens and data capture sheets. Please see appendix E for an example of the data capture sheets that were used.

3.5 Heparin measurement

Heparin levels were measured using an anti-Xa chromogenic assay. This test is based on a synthetic chromogenic substrate and on Factor Xa inactivation. It is used for the quantitative determination of unfractionated heparin and low molecular weight heparin activity in human citrated plasma. Washing of blood with a ATS removes most of the plasma proteins including clotting factors and AT III. At the start of this test, cell saver blood is supplemented with purified AT III. The heparin in the sample forms complexes with the AT III. A known amount of FXa is then added, which results in the formation of heparin-ATIII-FXa complexes. The excess FXa, which remains, releases the chromogen p-nitroanaline from a chromogenic substrate. The amount of p-nitroanaline is inversely proportional to the amount of heparin in the sample.(49)

Quality control: Controls were run before any number of batched tests were conducted. For calibration purposes a standard preparation of 0.8 units of heparin per millilitre was prepared using the same heparin used during CPB. Normal Pooled Plasma (NPP) was used in preparation of the standard. During the control sample testing, we consistently found results with almost half the value of the known concentrations prepared for control purposes. Similar problems have been encountered in studies employing heparin chromogenic tests in samples not resembling plasma. The study of heparan sulphate's anticoagulant activity in human follicular fluid by de Agostini et al(50) is one example. In this study the problem was solved by measuring the anti-Factor Xa level after making 1:2 to 1:16 dilutions in normal plasma and correcting for this dilution with the appropriate calculations. We have found that making 1:2 dilutions of our samples with normal pooled plasma resulted in control samples with reliable results.

3.6 Data storage

Results from the heparin analysis was accumulated and presented in Excel document format (Appendix A).

3.7 Ethical aspects

The study was approved by the Committee for Human Research of the University of Stellenbosch on the 08-08-2014 (Reference: S14/03/050, see appendix B). The research conducted adhered to the ethical guidelines and principles of the International Declaration of Helsinki, South African Guidelines for Good Clinical Practice and the Medical Research Council (MRC) Ethical Guidelines for Research. The aim of this study was to provide answers in improving the quality of care of patients receiving cell saver blood in our institution. There was no perverse incentive, and no investigator had any relationship, commercial or otherwise, with the company or its employees that supply the device. The study population was fairly chosen, and not particularly vulnerable. No groups that could benefit from the research were excluded. Patient autonomy was respected through invitation to join the study with a personal interview by the lead investigator preoperatively. Informed consent was taken during this interview and included an explanation of the study, its implications and possible benefits. Each patient was provided an information sheet that included the contact numbers of the ethical committee and investigator, should they have any queries (Appendix C,D). Patient identity was protected by de-identification of the samples using a numbering system during collection. The data sheets with the patient's identities were collected by the lead investigator and kept securely.

3.8 Resources

The Sorin Xtra® Cell Separator device is currently in use at Tygerberg Academic Hospital as standard practice for all cardiac related procedures requiring CPB. Written permission was obtained from the CEO (Dr Erasmus) of Tygerberg Hospital to perform the study. The modified Anti-Xa assay was performed by the same laboratory technologist involved in the first study (Ms Marieta du Plessis). The tests were performed by the National Health Laboratory Services (NHLS) on site at Tygerberg Hospital. Written permission was obtained from the NHLS to utilise the laboratory services and staff in support of this study. The account from the NHLS was settled by the Department of Anesthesiology, from their research fund. A R15000 research bursary from the Harry Croxley fund was utilised for this.

4. Results and discussion

31 patients were enrolled in this study. Mean reinfusate heparin levels measured from the Sorin® Xtra® ATS were 0.52 (interquartile range 0.16 to 0.74; 95% confidence interval 0.30 to 0.66) IU/ml(see Figure 1). The null hypothesis tested by a one-sample test against 0.5 U/mL was that the Sorin Xtra® reinfusate contained clinically significant heparin concentrations. The data failed to reject the null hypothesis and thus are consistent with clinically significant reinfusate heparin concentrations. Sixteen of 31 reinfusates (56%; 95% confidence interval 35 to 68%) exhibited heparin concentrations exceeding 0.5 IU/ml. There was no significant relationship between the heparin dosages administered to the patient and reinfusate heparin concentrations(r^2 0.02, p = 0.46).

The 2002 and subsequent American Association of Blood Banks' guidelines state that reinfusate heparin concentrations should be below 0.5 anti-Xa IU/ml.(46, 48, 51, 52) This is identical to Yawn and colleagues' recommendation, but slightly higher than Umlas and colleague's suggested maximal range of 0.2 to 0.4 IU/ml.(33, 34) The average reinfused heparin doses would be 418 IU (6.0 IU/kg for a 70 kg patient) representing very small doses of heparin compared to those typically administered (200 to 300 IU/kg) before initiation of cardiopulmonary bypass. Such small doses of heparin are likely inconsequential. We elected not to investigate either the effects of these small dosages of heparin or the deleterious effects of the reinfusate on the vulnerable coagulation status or bleeding following cardiac surgery. Not doing so does represent a limitation of our study. However, accounting for all possible above-mentioned reinfusate anticoagulant effects would be costly and complicated, point of care identification of low levels of residual heparin being difficult.(23, 53-57)

Our primary concern was whether the autotransfusion systems tested efficiently removed heparin, since reinfusate containing significant amounts of heparin could aggravate post cardiopulmonary bypass coagulopathy. Previous studies investigating cell saver reinfusates, have measured similar or even higher (0.64, 0.67 to 1.71, and 0.61 to 0.8 IU/ml) heparin concentrations than in our studies (Table 1).(40-43) To ensure reinfusate heparin concentrations were measured accurately, it is essential to report the measurement method used and to explicitly state that the critical

modification described above was employed. It is unclear whether this critical modification was indeed employed in any of the previous studies investigating reinfusate heparin concentrations (Table 1), nonconformity causing underestimation of reinfusate heparin concentrations.(11, 33, 35-47, 58-60)

The volume of heparin containing wash fluid did not affect reinfusate heparin concentrations. In one of the few studies examining device performance, differing cell saver wash regimens did not affect heparin elimination.(47) In this respect, a further limitation of our study was not studying the effect of different washing regimens (alterations of the number of wash cycles, volumes of saline used to wash the reinfusate, wash fluid heparin concentrations or bowl sizes) and reinfusate heparin concentrations. As some aging devices have performed poorly, periodic quality control of reinfusate composition, including heparin concentrations, has been recommended.

5. Conclusion

Clinically significant heparin levels in cell saving reinfusate can potentially worsen postoperative bleeding in cardiac surgery. The mean heparin level measured in our study was more than the AABB's recommended value of 0.5IU/ml, and 16 samples had absolute values more than this. Although the absolute heparin dose retransfused remain debatably low, the possibility of heparin induced coagulopathy should be entertained in cardiac patients that received reinfusate from the Sorin Xtra® ATS with ongoing postoperative bleeding in our institution. A practical suggestion in these cases would be to quantify heparin activity either with a point of care device (TEG/ROTEM) or direct measurement of heparin concentration using an Anti-Xa assay and titrating heparin reversal accordingly.

Tables

Table 1: Available literature investigating heparin removal in autotransfusion systems.

Author	Year	Autotransfusion device studied	Residual heparin in washed blood (reported as mean ± standard deviation unless stated otherwise)	Assay utilised	Conclusion
Umlas et al(33)	1981	Haemonetics Cell Saver	In vitro study	Grann assay (polybrene neutralization of heparin effect)	Not stated
Ottesen et al(59)	1982	Haemonetics Cell Saver	0.019 ± 0.02 IU/ml	Thrombin-protamine-heparin neutralization assay	"of no significance"
Paravicini et al(42)	1983	Haemonetics Cell Saver	Maximum heparin level in one unit of cell-saver blood was 60 IU	"a highly sensitive heparin test"	"totally free of heparin"
Sandmann et al(41)	1985	Haemonetics Cell saver 3 Rapid Autotransfusion Machine	0.16 IU/ml (IQR 0.0 – 0.46 IU/ml)	Not stated	Not stated

Mc Shane et al(58)	1987	Dideco Autotrans BT 795	0.64± 0.3 IU/ml	Not stated	"..modification of the centrifugation and washing is required to lessen the high white cell count and heparin concentrations found in the saved blood."
Kling et al(40)	1988	Haemonetics Cell Saver 4	0.41 (IQR 0.07-0.77) IU/ml	Anti-Xa chromogenic assay (antithrombin III added to samples but addition of pooled plasma not stated) [Testkombination Heparin, Boehringer Mannheim, Germany]	"amount of heparin is not enough to provoke bleeding"
Sistino et al(39)	1992	Haemonetics Cell Saver 4	0.0027 ± 0.03 IU/ml	Anti-Xa chromogenic assay (Modification by addition of pooled plasma not stated) (Stachrom, Medtronic Hemotec, Inc., Englewood, CO, USA)	"no clinical significance"
Gravlee et al(60)	1992	Haemonetics cell saver	<0.04 IU/ml with 750ml saline wash 0.08-0.22 IU/ml with 500ml saline wash	Whole blood heparin concentration (Hepcon, HemoTec Inc., Englewood, CO)	"usual complete wash cycle do not contain clinically significant amounts of heparin"
Rougé et al(38)	1993	Haemonetics Cell Saver 4 Dideco/Shiley STAT BRAT 250	0.269 ± 0.01 IU/ml 0.223 ± 0.05 IU/ml 0.463 ± 0.01 IU/ml	Anti-Xa chromogenic assay (Modification by addition of pooled plasma not stated) (Stachrom Heparin, Diagnostica Stago, Asnieres, France)	"virtual elimination of heparin"
Kalra et al(37)	1993	Haemocell System 350	4.93 ± 0.91 IU/ml 0.10 IU/ml mean plasma heparin level 4 hours postoperative	Anti-Xa chromogenic assay (Modification by addition of pooled plasma not stated) (Coatest Heparin kit, KabiVitrum B.V.Diagnostica, Amsterdam, The Netherlands)	"plasma heparin concentration was negligible"
Vorweg et al(36)	1998	Cell Saver 5	Measured range 0.00 – 2.44 IU/ml	["Heparin Test für ACS (Fa. Date-Behring)"]	"no irregular heparin load but, if the volume of rinsing liquid is decreased or the pumpflow is increased, the heparin load is increased enormously"

Geiger et al(43)	1998	Dideco Compact A Haemonetics Haemolite 3 Haemonetics Cell Saver 5 Fresenius C.A.T.S. Medtronic AutoLog Medtronic Sequestra Transf. Tech OrtoPAT	0.40 IU/ml 0.29 IU/ml 0.72 IU/ml 1.17 IU/ml 0.79 IU/ml 0.67 IU/ml 0.10 IUml (all values are median values)	Not stated	"very effective washout"
Levy et al(44)	2001	Dideco Electa 5.0	<0.1 IU/ml Heparin elimination rate 98.7 ± 0.2%	Not stated	"Efficiently removes heparin"
Cuby et al(45)	2001	Dideco Electa 5.0	<0.1 IU/ml Heparin elimination rate 98.7 ± 1.7%	Not stated	'Residual heparin levels low"
Burman et al(11)	2002	Cobe BRAT 2 Fresenius C.A.T.S. Medtronic Sequestra Haemonetics Cell Saver 5 Dideco Compact A	0.3 (measured range: 0 – 4.0) IU/ml	Anti-Xa chromogenic assay (Modification by addition of pooled plasma not stated) (Stachrom, Diagnostica Stago, Dundee, Scotland)	"Heparin was effectively removed"
Serrick et al(35)	2003	Cobe BRAT 2 Medtronic Sequestra 1000 Haemonetics Cell Saver 5 Medtronic Autolog Fresenius C.A.T.S.	0.80 ± 0.65 IU/ml 0.61 ± 0.36 IU/ml 0.19 ± 0.17 IU/ml 0.39 ± 0.33 IU/ml 0.73 ± 0.86 IU/ml	Anti-Xa chromogenic assay (Modification by addition of pooled plasma not stated)	"All devices adequately removed heparin"
Kelleher et al(46)	2011	Cobe BRAT 2 Haemonetics Cell Saver 5	BRAT 2 2001: 0.2 (0.0-0.4[0-1.9]) 2002: 0.2 (0.1-0.5[0-1.7]) 2003: 0.4 (0.2-2.3[0-1.7]) 2004: 0.3 (0.2-0.5[0-0.8]) 2005: 0.2 (0-0.4[0-1.7]) 2006:0.3 (0-0.5[0-0.6]) Cell Saver 5 2006: 0.4 (0.2-0.7[0-2.5]) 2007: 0 (0.0-0.2[0-0.4]) year: median (IQR[range])	Anti-Xa chromogenic assay (Modification by addition of pooled plasma not stated) (Stachrom, Diagnostica Stago, Asnieres, France)	"Consistent heparin removal"

Overdevest et al(47)	2012	Sorin Xtra®	Heparin elimination rate 98.8-99.9%	Anti-Xa chromogenic assay (Modification by addition of pooled plasma not stated) (Roche Stago)	removal rate, depending on wash set and wash speed "performs adequately"

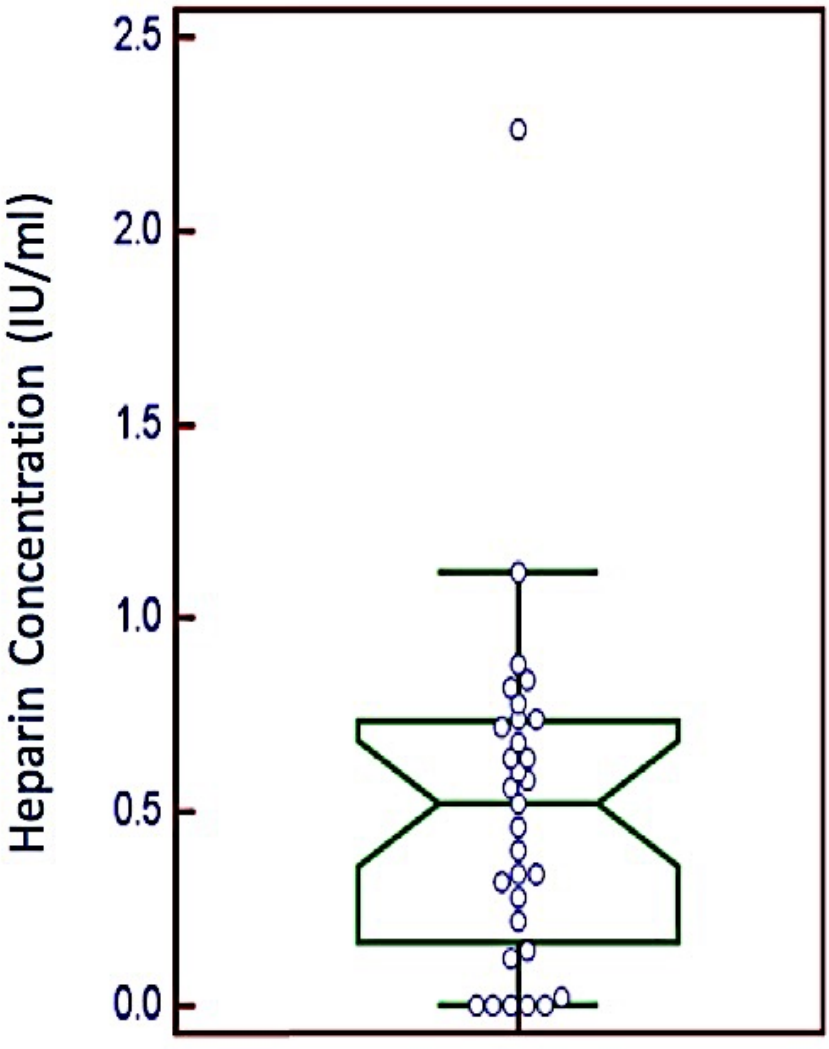
Notes: Year denotes the year the article was published. Note that Levy et al and Cuby et al's articles were only published as abstracts.

BRAT 2 and BRAT 250 (Cobe Cardiovascular, Quedgeley, UK); C.A.T.S. (Fresenius Kabi Ltd., Runcom, UK); Sequestra, Sequestra 1000, and AutoLog (Medtronic Ltd., Watford, UK); Cell Saver, Cell Saver 4, and Cell Saver 5 (Haemonetics, Braintree, MA, USA); Compact A, and Shiley STAT (Dideco, Sorin Biomedica UK, Quedgeley, UK); System 350 (Haemocell, Abington, UK); OrtoPAT (Transf. Tech, (Haemonetics), Braintree, MA, USA), Electa 5,0 (Dideco, Modena, Italy); Sorin Xtra® (Sorin Group, Mirandola, Italy); Autotrans BT 795 (Dideco S.p.A., Mirandola, Italy); Rapid Autotransfusion Machine (Baylor College of Medicine, Houston). Note that the Sorin Group has acquired the Cobe and Dideco companies.

Abbreviations: IQR: interquartile range.

Figures

Figure 1: Individual points and notched box-and-whisker plot of absolute heparin concentrations measured in reinfusate from Sorin Xtra® ATS. Mean heparin concentration 0.52 IU/ml; IQR 0.16 to 0.74 IU/ml; 95% confidence interval 0.30 to 0.66 IU/ml



Appendices

Appendix A: Excel spreadsheat with data

Test results: Sorin Xtra® Heparin in Cell Saver Blood				
			Specimen Registered	AF10A
Labno	Name	Locn	Date	AF10A
STY7066351	SAMPLE E1	X5147	20/05/2015	0.4
STY7067004	SAMPLE E3	X5147	20/05/2015	0.32
STY7067007	SAMPLE E4	X5147	20/05/2015	0.22
STY7067012	SAMPLE E5	X5147	20/05/2015	0.78
STY7067019	SAMPLE E6	X5147	20/05/2015	0.14
STY7067022	SAMPLE E02	X5147	20/05/2015	0.72
STY7067024	SAMPLE E8	X5147	20/05/2015	0.84
STY7067026	SAMPLE E9	X5147	20/05/2015	0.68
STY7067030	SAMPLE E10	X5147	20/05/2015	0.56
STY7067033	SAMPLE E11	X5147	20/05/2015	2.26
STY7067045	SAMPLE E11	X5147	20/05/2015	2.26
STY7067058	SAMPLE E14	X5147	20/05/2015	0.28
STY7067063	SAMPLE E15	X5147	20/05/2015	0.74
STY7067068	SAMPLE E16	X5147	20/05/2015	0.82
STY7067072	SAMPLE E17	X5147	20/05/2015	0.58
STY7067078	SAMPLE E18	X5147	20/05/2015	0.74
STY7067085	SAMPLE E19	X5147	20/05/2015	0
STY7067089	SAMPLE E21	X5147	20/05/2015	0
STY7067093	SAMPLE E22	X5147	20/05/2015	0
STY7067103	SAMPLE F2	X5147	20/05/2015	0.6
STY7067104	SAMPLE F3	X5147	20/05/2015	0.34
STY7067106	SAMPLE F4	X5147	20/05/2015	0.64
STY7067109	SAMPLE F5	X5147	20/05/2015	0
STY7067110	SAMPLE F6	X5147	20/05/2015	0.46
STY7067112	SAMPLE F7	X5147	20/05/2015	0.34
STY7067113	SAMPLE F8	X5147	20/05/2015	0.52
STY7067118	SAMPLE F9	X5147	20/05/2015	0.02
STY7067120	SAMPLE F10	X5147	20/05/2015	0.64
STY7067500	SAMPLE F11	X5147	20/05/2015	0.88
STY7067513	SAMPLE F12	X5147	20/05/2015	0
STY7067514	SAMPLE F13	X5147	20/05/2015	1.12
STY7067515	SAMPLE F14	X5147	20/05/2015	0.12

Appendix B: HREC acceptance letter

UNIVERSITEIT·STELLENBOSCH·UNIVERSITY
jou kennisvennoot · your knowledge partner

Approval Notice
New Application

08-Aug-2014
Buys, Mathilde M

Ethics Reference #: S14/03/050
Title: **Residual heparin concentrations in salvaged blood from the Sorin Xtra autotransfusion system during cardiac surgery.**

Dear Dr Mathilde Buys,

The **New Application** received on **17-Mar-2014**, was reviewed by members of **Health Research Ethics Committee 1** via Expedited review procedures on **04-Aug-2014** and was approved.
Please note the following information about your approved research protocol:

Protocol Approval Period: **04-Aug-2014 -04-Aug-2015**

Please remember to use your **protocol number** (**S14/03/050**) on any documents or correspondence with the HREC concerning your research protocol.

Please note that the HREC has the prerogative and authority to ask further questions, seek additional information, require further modifications, or monitor the conduct of your research and the consent process.

After Ethical Review:
Please note a template of the progress report is obtainable on www.sun.ac.za/rds and should be submitted to the Committee before the year has expired. The Committee will then consider the continuation of the project for a further year (if necessary). Annually a number of projects may be selected randomly for an external audit.
Translation of the consent document to the language applicable to the study participants should be submitted.

Federal Wide Assurance Number: 00001372
Institutional Review Board (IRB) Number: IRB0005239

The Health Research Ethics Committee complies with the SA National Health Act No.61 2003 as it pertains to health research and the United States Code of Federal Regulations Title 45 Part 46. This committee abides by the ethical norms and principles for research, established by the Declaration of Helsinki, the South African Medical Research Council Guidelines as well as the Guidelines for Ethical Research: Principles Structures and Processes 2004 (Department of Health).

Provincial and City of Cape Town Approval

Please note that for research at a primary or secondary healthcare facility permission must still be obtained from the relevant authorities (Western Cape Department of Health and/or City Health) to conduct the research as stated in the protocol. Contact persons are Ms Claudette Abrahams at Western Cape Department of Health (healthres@pgwc.gov.za Tel: +27 21 483 9907) and Dr Helene Visser at City Health (Helene.Visser@capetown.gov.za Tel: +27 21 400 3981). Research that will be conducted at any tertiary academic institution requires approval from the relevant hospital manager. Ethics approval is required BEFORE approval can be obtained from these health authorities.

We wish you the best as you conduct your research.
For standard HREC forms and documents please visit: www.sun.ac.za/rds

If you have any questions or need further assistance, please contact the HREC office at 0219389657.

Included Documents:
Budget
Investigator declaration du Plessis
Investigator declaration WFBuys
CV du Plessis

Appendix C: Consent form and information leaflet (English)

PARTICIPANT INFORMATION LEAFLET AND CONSENT FORM

Title of the Research Project: Residual heparin concentration in cell saver blood from the Sorin Xtra® autotransfusion system.

Principal Investigator: Dr Mathilde Buys, Department of Anaesthesiology and Critical Care, Tygerberg Academic Hospital. CONTACT NUMBER: 021 9385142 or 0716059345

Introduction to a research project: your rights and responsibilities:

1. You are being invited to take part in a research project.
2. Please take some time to read the information presented here, which will explain the details of this project.
3. Please ask the study doctor any questions about any part of this project that you do not fully understand. It is very important that you are fully satisfied that you clearly understand what this research entails and how you could be involved.
4. Also, note that your participation is **entirely voluntary** and you are free to decline to participate.
5. If you say no, this will not affect you negatively in any way whatsoever. You are also free to withdraw from the study at any point, even if you do agree to take part.
6. Please note that the Ethical and scientific aspects of this study have been carefully looked at by the Ethics committee for your protection. This study has been approved by the **Committee for Human Research at Stellenbosch University** (The Ethics committee) and will be conducted according to the ethical guidelines and principles of the international Declaration of Helsinki, South African Guidelines for Good Clinical Practice and the Medical Research Council (MRC) Ethical Guidelines for Research.

What is this research study all about?

1. This study will be conducted in the heart theatres at Tygerberg Academic Hospital.
2. Approximately 29 patients will be studied.
3. A standard operation will be performed.
4. As part of the ordinary running of heart operations, any of your own blood that is lost into the heart-lung machine is to be returned to you using a special blood conservation device. This is done to minimise the use of blood from the blood bank in order to limit the side effects associated with banked blood.
5. This device washes your own blood before it is given back to you.
6. The purpose of this project is to **check the quality of the washed blood**. Specifically, we are interested in determining the amount of heparin (blood thinner or anticoagulant) remaining in the blood.
7. The project involves taking a small sample (approximately a teaspoon) of your processed blood for testing before we give the blood back to you. The quality of the blood will not be changed in any way.

Why have you been invited to participate?

This project involves 29 patients undergoing elective heart operations like yourself.

What will your responsibilities be?

You have no additional responsibilities. Your operation will be conducted as usual.

Will you benefit from taking part in this research?

1. This project aims at testing the quality of the blood product that we return to patients.
2. There will be no immediate benefit during your operation.
3. Future patients will benefit from this research.

Are there any risks involved in your taking part in this research?

There are no additional risks involved if you take part in this project.

What will happen in the unlikely event of some form of injury occurring as a direct result of your taking part in this research study?

1. It is extremely unlikely that injury can occur as a direct result of your taking part in this research project.
2. In the extremely unlikely event of injury, the University has insurance to cover such eventualities. Such cases will be evaluated according to Association of British Pharmaceutical Industry compensation guidelines.

If you do not agree to take part, what alternatives do you have?

Your operation will be conducted in exactly the same way whether you decide to participate in this project or not.

Who will have access to your medical records?

1. The information collected will be treated as confidential and protected.
2. If it is used in a publication, your identity will remain anonymous.
3. Only the investigators involved in this project will have access to your information.

Will you be paid to take part in this study and are there any costs involved?

1. No you will not be paid to take part in the study.
2. There will be no costs involved for you, if you do take part.

Is there anything else that you should know or do?

1. You can contact Dr Mathilde Buys at telephone 0716059345 if you have any further queries or encounter any problems regarding the study.
2. You can contact the Committee for Human Research at 021-938 9207 if you have any concerns or complaints that have not been adequately addressed by your study doctor.

3. You will receive a copy of this information and consent form for your own records.

Declaration by participant

By signing below, I ……..……. agree to take part in a research study entitled **Residual heparin concentration in cell saver blood from the Sorin Xtra® autotransfusion system.**

I declare that:

1. I have read or had read to me this information and consent form.
2. It is written in a language with which I am fluent and comfortable.
3. I have had a chance to ask questions and all my questions have been adequately answered.
4. I understand that taking part in this study is **voluntary** and I have not been pressurised to take part.
5. I may choose to leave the study at any time and will not be penalised or prejudiced in any way.

Signed at (place) **Tygerberg Academic Hospital** on (date) ……...…….. .....

....

Signature of participant **Signature of witness**

Declaration by investigator

I (name) ……..…… declare that:

1. I explained the information in this document to ……..
2. I encouraged him/her to ask questions and took adequate time to answer them.
3. I am satisfied that he/she adequately understands all aspects of the research, as discussed above.
4. I did/did not use an interpreter. (If an interpreter is used then the interpreter must sign the declaration below.

Signed at (place) **Tygerberg Academic Hospital** on (date) ……...…….....

....

Signature of investigator **Signature of witness**

Declaration by interpreter

I (name) ……..…… declare that:

1. I assisted the investigator (name) ……. to explain the information in this document to (name of participant) ……..…….. using the language medium of Afrikaans/Xhosa.
2. We encouraged him/her to ask questions and took adequate time to answer them.
3. I conveyed a factually correct version of what was related to me.
4. I am satisfied that the participant fully understands the content of this informed consent document and has had all his/her question satisfactorily answered.

Signed at (place) **Tygerberg Academic Hospital** on (date) ……...……..

....

Signature of interpreter **Signature of witness**

Appendix D: Informed consent and information leaflet (Afrikaans)

DEELNEMERINLIGTINGSBLAD EN -TOESTEMMINGSVORM

Titel van die Navorsingsprojek: Oorblywende heparien in sel-besparings bloed met die gebruik van die Sorin Xtra® autotransfusie toestel.

Hoofnavorser: Dr Mathilde Buys, Departement Anestesiologie en Kritieke Sorg, Tygerberg Akademiese Hospitaal. KONTAKNOMMER: 021 9385142 of 0716059345

Inleiding tot navorsingsprojek: u regte en verantwoordelikhede:

1. U word uitgenooi om deel te neem aan ‘n navorsingsprojek.
2. Lees asseblief hierdie inligtingsblad op u tyd deur aangesien die detail van die navorsingsprojek daarin verduidelik word.
3. Indien daar enige deel van die navorsingsprojek is wat u nie ten volle verstaan nie, is u welkom om die navorsingsdokter daaroor uit te vra.
4. Dit is baie belangrik dat u ten volle moet verstaan wat die navorsingsprojek behels en hoe u daarby betrokke kan wees.
5. U deelname is ook **volkome vrywillig** en dit staan u vry om deelname te weier.
6. U sal op geen wyse hoegenaamd negatief beïnvloed word indien u sou weier om deel te neem nie. U mag ook te eniger tyd aan die navorsingsprojek onttrek, selfs al het u ingestem om deel te neem. **Hierdie navorsingsprojek is deur die** Komitee vir Mensnavorsing van die Universiteit Stellenbosch **goedgekeur en sal uitgevoer word volgens die etiese riglyne en beginsels van die Internasionale Verklaring van Helsinki en die Etiese Riglyne vir Navorsing van die Mediese Navorsingsraad (MNR).**

Wat behels hierdie navorsingsprojek?

1. Hierdie projek word slegs by Tygerberg Akademiese Hospitaal in die hartteaters uitgevoer.
2. Nagenoeg 29 pasiënte sal gewerf word vir die projek.

3. ‘n Standaard operasie sal uitgevoer word.
4. In die uitvoering van ‘n hartoperasie word enige van u eie bloed wat verlore gaan in die hartlongmasjien aan u terugbesorg deur middel van ‘n spesiale bloedbesparingstoestel. Dit word gedoen om die gebruik van bloed vanaf die bloedbank te verminder en sodoende die newe-effekte geassosieer met die toediening van bloed vanuit die bloedbank te beperk.
5. Die toestel was u eie bloed voor dit aan u terugbesorg word.
6. Die doel van hierdie projek is om die kwaliteit van die gewasde bloed na te gaan. Ons is veral geinteresseerd in die hoeveelheid heparien (bloedverdunner of anti-stolmiddel) wat oorbly in die bloed na die wasproses.
7. Die projek behels die neem van ‘n bietjie (omtrent ‘n teelepel) van u geprosesseerde bloed vir ontleding voordat die bloed weer aan u oorgetap word. Die bloed sal op geen manier verander of geaffekteer word nie.

Waarom is u genooi om deel te neem aan hierdie projek?

Die projek betrek 29 pasiënte wat elektiewe hartoperasies ondergaan soos uself.

Wat sal u verantwoordelikhede wees?

U het geen addisionele verantwoordelikhede nie. U operasie word op dieselfde manier uitgevoer ongeag u deelname aan hierdie studie aldan nie.

Sal u voordeel trek deur deel te neem aan hierdie navorsingsprojek?

1. Hierdie projek beoog om die kwaliteit van die genoemde bloedproduk na te gaan en te verseker.
2. Daar is geen onmiddelike voordeel tydens u operasie nie.
3. Toekomstige pasiënte sal baatvind by hierdie projek.

Is daar enige risiko's verbonde aan u deelname aan hierdie navorsingsprojek?

Daar is geen addisionele risiko’s verbonde aan u deelname aan hierdie projek nie.

Wat sal gebeur in die onwaarskynlike geval van 'n besering wat mag voorkom as gevolg van u deelname aan hiedie navorsingsprojek?

1. Dit is hoogs onwaarskynlik dat enige besering mag voorkom as 'n direkte gevolg van u deelname aan hierdie navorsingsprojek.
2. In die hoogs onwaarskynlike geval van besering, het die Universiteit assuransie om sulke gebeurlikhede te dek. Enige sulke gevalle sal ge-evalueer word volgens die *Association of British Pharmaceutical Industry*-vergoedingsriglyne.

Watter alternatiewe is daar indien u nie instem om deel te neem nie?

U operasie word op dieselfde manier uitgevoer ongeag u deelname aan hierdie studie.

Wie sal toegang hê tot u mediese rekords?

1. Die inligting wat versamel word, word vertroulik en beskermd hanteer.
2. Die deelnemer sal anoniem bly indien dit gebruik sou word vir 'n publikasie.
3. Slegs die navorsers wat betrokke is by die studie sal toegang tot die inligting hê.

Sal u betaal word vir deelname aan die navorsingsprojek en is daar enige koste verbonde aan deelname?

1. U word nie betaal vir deelname aan die navorsingsprojek nie.
2. Deelname aan die navorsingsprojek sal u niks kos nie.

Is daar enigiets anders wat u moet weet of doen?

1. U kan Dr Mathilde Buys kontak by tel 0716059345 indien u enige verdere vrae het of enige probleme ondervind.
2. U kan die Komitee vir Mensnavorsing kontak by 021-938 9207 indien u enige bekommernis of klagte het wat nie bevredigend deur u studiedokter hanteer is nie.

3. U sal ’n afskrif van hierdie inligtings- en toestemmingsvorm ontvang vir u eie rekords.

Verklaring deur deelnemer

Met die ondertekening van hierdie dokument onderneem ek, …….....……...……...……., om deel te neem aan ’n navorsingsprojek getiteld: **Oorblywende heparien in sel-besparings bloed met die gebruik van die Dideco Electa Cell Separator toestel.**

Ek verklaar dat:

1. Ek hierdie inligtings- en toestemmingsvorm gelees het of aan my laat voorlees het en dat dit in ’n taal geskryf is waarin ek vaardig en gemaklik mee is.
2. Ek geleentheid gehad het om vrae te stel en dat al my vrae bevredigend beantwoord is.
3. Ek verstaan dat deelname aan hierdie navorsingsprojek **vrywillig** is en dat daar geen druk op my geplaas is om deel te neem nie.
4. Ek te eniger tyd aan die navorsingsprojek mag onttrek en dat ek nie op enige wyse daardeur benadeel sal word nie.

Geteken te *(plek)* **Tygerberg Akademiese Hospitaal** op *(datum)* ……...……..

....

Handtekening van deelnemer **Handtekening van getuie**

Verklaring deur navorser

Ek *(naam)* ……...…… verklaar dat:

1. Ek die inligting in hierdie dokument verduidelik het aan …….....……..
2. Ek hom/haar aangemoedig het om vrae te vra en voldoende tyd gebruik het om dit te beantwoord.
3. Ek tevrede is dat hy/sy al die aspekte van die navorsingsprojek soos hierbo bespreek, voldoende verstaan.
4. Ek 'n tolk gebruik het/nie 'n tolk gebruik het nie. (*Indien 'n tolk gebruik is, moet die tolk die onderstaande verklaring teken.)*

Geteken te *(plek*) **Tygerberg Akademiese Hospitaal** op *(datum*) ……...……...

....

Handtekening van navorser **Handtekening van getuie**

Verklaring deur tolk

Ek *(naam)* ……...…… verklaar dat:

1. Ek die navorser *(naam*) …….....……. bygestaan het om die inligting in hierdie dokument in Afrikaans/Xhosa aan *(naam van deelnemer)* ……..... te verduidelik.

2. Ons hom/haar aangemoedig het om vrae te vra en voldoende tyd gebruik het om dit te beantwoord.
3. Ek ’n feitelik korrekte weergawe oorgedra het van wat aan my vertel is.
4. Ek tevrede is dat die deelnemer die inhoud van hierdie dokument ten volle verstaan en dat al sy/haar vrae bevredigend beantwoord is.

Geteken te *(plek)* **Tygerberg Akademiese Hospitaal** op *(datum)* ……...……..

....

Handtekening van tolk **Handtekening van getuie**

Appendix D: Data capture sheet (example)

Heparin in Cell Saver Blood: Theatre Report Form

Demographic Information/ Hospital label	
Patient identifier:	Group: Electa / Xtra
Date: / /	Weight: kg
Age:	Height: cm
Gender:	Procedure:

Cell Saver Use (info on cell saver screen)	
Processed volume	
Collected volume	
Bowls	
Reservoir volume	
Time	
Hematocrit	

Heparin information	
Patient: Total heparin administered by anaesthesiologist	
Technologist: Total heparin administered - pump technologist (excluding cell saver)	
Cell saver: Total volume of heparin wash bag administered	
Transducer flush: Bag one: 200 ml with 500 IU heparin: total volume:	¼ / ½ / ¾ / all
Transducer flush: Bag two: 200 ml with 500 IU heparin: total volume:	¼ / ½ / ¾ / all
Volume of pericardial and pleural blood loss in the first 12 hours post-op	

Bibliography

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